



## Effects of land management practices on water quality in Mississippi Delta oxbow lakes: Biochemical and microbiological aspects

R.M. Zablotowicz<sup>a</sup>, P.V. Zimba<sup>b</sup>, M.A. Locke<sup>c</sup>, S.S. Knight<sup>c</sup>,  
R.E. Lizotte Jr.<sup>c,\*</sup>, R.E. Gordon<sup>a</sup>

<sup>a</sup> USDA-ARS, Crop Production Systems Research Unit 141 Experiment Station Road, Stoneville, MS 38776, USA

<sup>b</sup> Center for Coastal Studies, Texas A&M University-Corpus Christi, Natural Resources Center 3200, 6300 Ocean Drive, Unit 5866, Corpus Christi, TX 78412, USA

<sup>c</sup> USDA-ARS, Water Quality & Ecology Research Unit, National Sedimentation Laboratory, P.O. Box 1157, Oxford, MS 38655, USA

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### ABSTRACT

The Mississippi Delta Management Systems Evaluation Area (MSEA) project was designed to assess the effects of land management practices on water quality in three small oxbow lake watersheds; Thighman (1338 ha, 16 ha lake); Beasley (915 ha, 25 ha lake); and Deep Hollow (132 ha, 8 ha lake). Monthly water samples were monitored for enzymatic activity (fluorescein diacetate hydrolysis, alkaline phosphatase, and substrate utilization), chemical and physical analysis (suspended solids, dissolved organic carbon, pH, nitrate, ammonium, orthophosphate, and electrical conductivity), phytoplankton and bacterioplankton populations. All of these parameters were influenced by the intrinsic nature of the watersheds, with some parameters shifting as management changes were imposed on the surrounding agricultural fields. Thighman lake water typically maintained the highest suspended solid levels, dissolved organic carbon, algal and bacterial populations, enzyme activities, and heterotrophic metabolic indexes. Introduction of reduced tillage practices and glyphosate-resistant crops in Beasley watershed resulted in lower levels of suspended sediments, but had minimal impact on overall ranking of biochemical or microbiological properties. Likewise, conversion of Deep Hollow watershed from reduced tillage to conventional tillage had little effect on suspended sediment, and most microbial activity parameters remained intermediate. However, canonical analysis indicated dynamic changes in the microbial community, suggesting that biological parameters of lake water quality were affected by changes in crop and soil management practices.

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### 1. Introduction

The Mississippi Delta is an area of intensive agricultural production and the impact of agricultural management on water quality is of great concern. The Mississippi Delta Management System Evaluation Area (MD-MSEA) project was established in 1994 as a collaborative project conducted by scientists from the US Department of Agriculture-Agricultural Research Service, US Geological Survey, and Mississippi State University, and supported by various federal, state and local agencies (Locke, 2004). The warm humid climate dictates extensive use of fertilizers and pesticides to maximize crop production, especially cotton (*Gossypium hirsutum* L.). MSEA projects have been established in several areas of the Midwestern USA (Ward et al., 1994) and have provided critical information for

assessing the effects of management practices on water quality. A major difference between the MD-MSEA and Midwestern MSEA projects is that the Mississippi Delta project was based on small watersheds that drain into oxbow lakes, while the Midwestern projects focused on watersheds that drain into creeks and streams. A second distinction is that the Mississippi project was designed to focus on watersheds with a majority of the acreage under a cotton cropping system, while the Midwestern project was under corn (*Zea mays* L.) and soybean (*Glycine max* [L.] Merr.) crop systems.

The microbial ecology of oxbow lakes is poorly understood, thus various analysis of biochemical characteristics, bacterioplankton and phytoplanktonic characterizations were conducted. In subtropical lakes, algal biomass typically is maximal during summer – due to large increases in cyanobacteria relative to green algae (Stchapit et al., 2008; Das and Chakrabarty, 2006). Initial data on the microbial ecology of the MD-MSEA lakes focusing on bacterial populations have been reported elsewhere (Zablotowicz et al., 2004). This study will report on findings from the second part of the study that was conducted

\* Corresponding author at: USDA-ARS National Sedimentation Laboratory, P.O. Box 1157, Oxford, MS 38655, USA. Tel.: +1 662 281 5703; fax: +1 662 232 2988.

E-mail address: [richard.lizotte@ars.usda.gov](mailto:richard.lizotte@ars.usda.gov) (R.E. Lizotte Jr.).

from 2000 to 2003 to ascertain planktonic response (using diagnostic photopigment analysis) to altered watershed management.

### 1.1. Area description

Three small oxbow lake watersheds in the central Mississippi Delta were selected as study areas. In order to maintain a watershed system framework and still address different kinds of practices, this project was designed so that each of the three oxbow lake watersheds was cropped and managed using a different set of Best Management Practices (BMPs), in a hierarchy ranging from no BMPs (Thighman Lake), edge-of-field practices only in Beasley, to a combination of edge-of-field and agronomic measures for conserving and improving soil and water resources (Deep Hollow Lake). A brief description of the watersheds is given herein, but details on physical characteristics, land management practices, and cropping patterns can be found elsewhere (Locke, 2004; Zablotowicz et al., 2006; Locke et al., 2008).

Thighman Lake watershed, Sunflower County, MS, is the largest of the three watersheds (drainage area 1499 ha), and has the smallest surface area of the three oxbow lakes (8.9 ha). Thus, runoff is concentrated into a fairly small water body. Thighman Lake watershed was designated as the program control, with all management decisions made by the farmers and no improvements initiated by the MSEA project. This watershed is the most diverse in terms of agricultural use and is managed by several farmers. Originally cotton was the dominant crop, however after 2000, less than 14% of the arable land was under cotton with a greater diversification in corn and soybeans. Although Thighman Lake watershed was the control, some of the farmers implemented conservation measures such as reduced tillage and cover crops during the study period. Overfall pipes and pads (berms) were installed in strategic locations in 2001 and 2002.

Thighman Lake riparian area comprises approximately 2% of the total watershed (27 ha). The largest riparian zone is bottomland hardwood deciduous forest with most of the vegetation abutting the north side of the lake. Riparian vegetation is comprised predominantly of bald cypress (*Taxodium distichum*) and oak (*Quercus* sp.). A narrow strip of riparian vegetation, 5–20 m wide, runs along the east side of the lake, while an intermittent strip of riparian vegetation, 0–10 m wide, occurs along the west side of the lake.

Beasley Lake watershed in Sunflower County, MS, has a drainage area of approximately 915 ha and has the largest surface area of the three oxbow lakes (25 ha). The Big Sunflower River defines the northern boundary of the watershed, and a large riparian-forested wetland (125–145 ha) is located on the eastern side of the lake. Beasley watershed was cropped predominantly in cotton and soybeans until 2002, with limited acreage in corn and sorghum. Structural and vegetative edge-of-field BMPs were established at Beasley Lake watershed from 1994 to 1996. Otherwise, agricultural management was left to the discretion of the farmers in the watershed. The structural BMPs included slotted board risers, and slotted inlet pipes, and vegetative filter strips established along the lake and in turn rows. The cropped area was maintained under conventional tillage until the spring of 2001, when glyphosate-resistant cotton and soybean facilitated the use of reduced tillage in much of the watershed. Beginning in spring 2003, 113 ha were removed from upland row crop production and planted in hardwood trees under the Natural Resources Conservation Service Conservation Reserve Program.

The Beasley Lake riparian area comprises approximately 15% of the total watershed. Riparian vegetation in Beasley is bottomland hardwood deciduous forest, predominantly oak. An extensive continuous riparian wetland is located in the eastern portion of the watershed, however, only about 20 ha directly abuts the east-

ern side of the lake. Another smaller riparian wetland abuts the northwestern side of the lake. A narrow band of continuous riparian vegetation, 10–30 m wide with predominantly bald cypress and oak, surrounds the entire lake.

Deep Hollow Lake watershed in Leflore County, MS, is the smallest of the three watersheds (drainage area of 203 ha) and has an 8.9-ha oxbow lake. The watershed is bordered by the Yazoo River on the west and has a large forested riparian zone on the western side of the lake. Both edge-of-field (structural and vegetative) and agronomic BMPs were established in Deep Hollow watershed. The land was managed by only one farmer, and throughout the study period, cotton was planted on the coarser-textured soils and soybeans on the heavier soils. A winter wheat cover crop was planted on all cropped land and was herbicide-desiccated prior to planting until the fall of 1999. Roundup Ready soybeans were grown using no-till management. Cotton was managed under minimum till with subsoiling and bed formation in the fall from 1994 to 2000. From the spring of 2000 until the end of the study all cotton and soybean land was returned to conventional tillage with several pre-plant tillage operations.

The Deep Hollow Lake riparian area comprises approximately 27% of the total watershed (36 ha). Riparian vegetation is predominantly oak with a few bald cypress trees. The largest continuous riparian wetland area abuts the west side of the lake. A narrow corridor of continuous riparian vegetation, 20–50 m wide with predominantly oak, surrounds the entire lake.

Cotton, soybean, and corn were the three major crops grown in the MD-MSEA watersheds during the years covered in this paper. Typical conventional tillage in Mississippi Delta row crops consists of plowing or disking in the fall, sometimes accompanied by subsoiling. Fields are then disked in the spring and row beds are formed. Several cultivations are included after crop emergence for weed control. Distinctions between “conventional tillage” and “reduced tillage” primarily refer to number of tillage operations as well as intensity of tillage. Reduced tillage usually involves disking once in either the fall or spring, shaping the beds, and planting. Also, reduced tillage, especially with the advent of herbicide-resistant crops, consists of less cultivation operations. Soybeans are often planted in narrow (less than 76 cm) rows, while corn and cotton rows are typically 1 m wide. Herbicides are applied to the soil as pre-emergence in all three crops, and with the herbicide-resistant systems, post-emergence herbicide applications are made to the crop canopy. Cultivation or post-emergence herbicide applications are normally performed until the crop is too high for equipment to move through the field.

## 2. Materials and methods

### 2.1. Water sample processing

Surface water samples were collected monthly from three permanent sampling rafts located in each of the three oxbow lakes as described elsewhere (Knight and Welch, 2004; Zablotowicz et al., 2004) from February of 2000 to December 2003. Samples were refrigerated immediately and stored at 5°C in the dark until processing for biological properties, typically within 24 h. At each site within each lake, standard water quality measurements of temperature (°C), pH (standard units), dissolved oxygen (mg L<sup>-1</sup>), and conductivity (μS cm<sup>-1</sup>) were collected *in situ* weekly during 2000 and every 2 weeks during 2001–2003. Measurements were determined using a calibrated Yellow Springs Instruments, Inc. (YSI) 556 multi-probe system instrument (Yellow Springs, OH) conforming to standard field methods (APHA, 2005). In addition, one (1) liter aqueous (water) samples were collected, preserved (via ice) and transported to the USDA-ARS

National Sedimentation Laboratory, Oxford, MS for nutrient analyses.

Alkaline phosphatase has been studied as a parameter of water quality in estuaries (Boyer, 2006). Alkaline phosphatase activity was determined using p-nitrophenol phosphate as substrate as described elsewhere (Locke et al., 2008). Heterotrophic biological activity of water samples was assessed by determining fluorescein diacetate (FDA) hydrolytic activity (Zablotowicz et al., 2001, 2004). Both alkaline phosphatase and FDA-hydrolytic activity were determined on all 47 monthly samples during the study. Multiple substrate utilization potential was determined using Biolog GN plates (Biolog, Hayward, CA). Water samples were diluted 100-fold and 150  $\mu$ L pipetted into each well, two replicate plates per water sample. After 72 h incubation at 25 °C, optical densities were analyzed using a Biotek plate reader at 485 nm. The average of well development for all 95 substrates were corrected for the no-substrate blank and analyzed as total average well development. Biolog substrate assays were initiated in April 2000 and conducted on alternate months.

Bacterioplankton populations (total heterotrophic bacteria and Gram-negative bacteria) were assayed by serial dilution and spiral plating as described elsewhere (Zablotowicz et al., 2001, 2004). Coliform populations were estimated using a most-probable-number (MPN) technique using five replicate tubes and BP-lauryl tryptose broth as media, positive tubes were scored based on gas production and acidification. Production of both acid and gas were used as indicators for tubes scored as positive. Algal populations were estimated using a five tube MPN and Bristol's mineral salts broth as media (Zablotowicz et al., 2001, 2004). Algal MPN tubes were incubated at 20 °C under low light (75  $\mu$ Einsteins  $m^{-2} s^{-1}$ ) for 1 month prior to evaluating for growth. Bacterioplankton and coliform assays were conducted on alternate month samples, algal MPN assays were conducted on alternate months in 2000 and on all monthly samples thereafter.

Phytoplankton community analysis was determined by measurement of diagnostic pigment biomarkers using high pressure liquid chromatography pigment analysis (Schlüter et al., 2006). Briefly, 500 mL of Beasley water and 250 mL of Deep Hollow and Thighman water were collected on GF/C filters (Whatman Corporation, Maidstone, England) in the dark. Filters were immediately frozen (−80 °C) until processed. Filters were extracted in 100% acetone after 30 s ultrasound disruption, clarified, then analyzed for carotenoid and chlorophyll content using a HP1100 high performance liquid chromatograph equipped with DAD detector (HP, Palo Alto, CA). Pigments were identified and quantified using authentic standards (VKI, Denmark) as previously described (Zimba et al., 2002). Using this technique pigments observed include: alloxanthin, aphanixanthin, chlorophyll *a*, chlorophyll *b*, chlorophyll *c2*, *b*-carotene, diadinoxanthin, fucoxanthin, lutein, neoxanthin, violaxanthin, and zeaxanthin.

## 2.2. Chemical and physical analysis

For determination of suspended solids, water samples (a total of 500 mL) were centrifuged in 250 mL polypropylene centrifuge bottles (10 min, 6000  $\times$  g). The supernatant was decanted, and the mass of the pellet determined following drying at 60 °C for 48 h. Electrical conductivity (EC) and pH was determined using appropriate electrodes. Total dissolved organic carbon (DOC) content was determined on water samples initially filtered through a 2  $\mu$ m filter and acidified (100  $\mu$ L 1.0 M HCl per 10 mL) to remove carbonates. A Flash EA 112 elemental analyzer (CE Elantech, Lakewood, NJ, USA) with an automated sampler delivering a 100  $\mu$ L injection was used with analysis and conducted in triplicate. Aspartic acid was used as a carbon and nitrogen standard. In most water samples from Beasley Lake and Deep Hollow Lake, total nitro-

gen content was below the limits of detection (1.0 mg  $L^{-1}$ ) for the instrumentation. Samples were analyzed for  $NO_3-N$ ,  $NH_4-N$ , and filterable orthophosphate (FOP) were filtered using a 0.45  $\mu$ m membrane filter according to APHA (2005). Inorganic nitrogen analyses, total nitrate-nitrogen ( $NO_3-N$ ) was determined via the cadmium reduction method and total ammonium-nitrogen ( $NH_4-N$ ) was determined using the phenate method (APHA, 2005). Nutrient analyses also included total orthophosphate (TOP). The ascorbic acid method was used for FOP analysis and TOP was determined by initial persulfate digestion followed by the ascorbic acid method (APHA, 2005).

## 2.3. Statistical analysis

Several approaches were utilized to compare the biochemical and microbiological properties of the three lakes and the effects of these interacting factors on lake water ecology. First, analysis of variance (ANOVA) was conducted for all parameters among the three lakes on a yearly basis, using PROC GLM of SAS (SAS, 2001). Canonical discriminant analysis was conducted using PROC CANDISC on data that was log [10] or log [10] + 1 transformed prior to analyses. Data were analyzed by lake, and standardized canonical coefficients were used to determine dominant variables contributing to yearly changes in lake structure.

## 3. Results and discussion

### 3.1. Lake chemical and physical properties

The average monthly total solids and electrical conductivity for the three lakes are presented in Fig. 1. The general pattern observed for highest total solids was that the highest densities were found

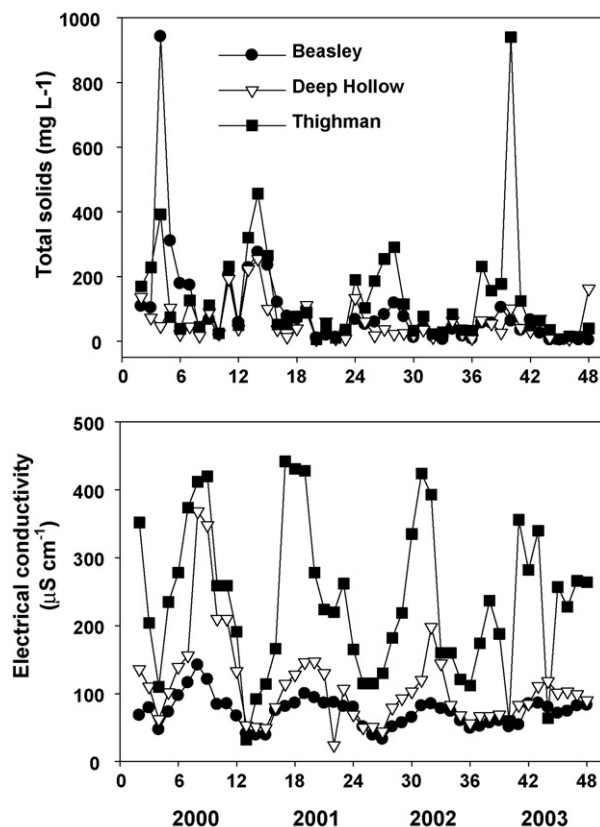


Fig. 1. Monthly mean total solids and electrical conductivity of three Mississippi Delta oxbow lakes, 2000–2003, each symbol represents the mean of three replicates.

in the spring months coinciding with the highest rainfall (February through May), while the highest electrical conductivity was observed in the summer months (May through August) with the warmest temperatures and lowest rainfall. In Beasley Lake the highest levels of suspended solids were found in 2000 and the spring of 2001, with lower levels thereafter reflecting a change in watershed management to more land under conservation tillage ( $p < 0.001$ ). Beasley Lake had maintained the highest levels of suspended solids compared to the other lakes from 1997 to 1999 (Knight and Welch, 2004; Zablotowicz et al., 2004). It should be noted that 2002 and 2003 had less rainfall, and lower sediment in these years may also be attributable to less runoff. Although Deep Hollow watershed was returned to conventional tillage in 2001 after 6 years of no-tillage, there were limited effects on suspended solid levels in that lake. This observation is supported by small plot rainfall simulations that indicated benefits of reduced tillage history on lower sediment loss despite recent tillage (Dabney et al., 2004; Krutz et al., 2009; Wilson et al., 2004). The highest EC values were typically observed in Thighman Lake with similar EC in Beasley and Deep Hollow Lake. However, a change of no-tillage to conventional tillage in Deep Hollow resulted in significant reductions in EC comparing levels observed in 2000 for the subsequent years (2001–2003;  $p < 0.01$ ).

Dissolved organic carbon (DOC) ranged from 3 to 45 mg CL<sup>-1</sup> with highly variable concentrations throughout the 4 year study (Fig. 2). Except for 2002, DOC was typically greatest in Thighman Lake, intermediate in Deep Hollow Lake and lowest in Beasley Lake (Table 1). The spike in DOC levels in Beasley Lake and Deep Hollow Lake water observed during August 2002 (month 32), accounted for no difference among the mean DOC levels in the lakes in 2002. No changes in DOC were observed with changes in land management in either Deep Hollow or Beasley Lake, while DOC in Thighman Lake

**Table 1**

Yearly mean of suspended solids, total organic carbon content, pH, and EC of water samples collected from three MSEA Mississippi Delta oxbow lakes (2000,  $n = 11$ ; 2002–2003,  $n = 12$ ).

Lake	2000	2001	2002	2003
Suspended solids (mg L <sup>-1</sup> )				
Beasley	200 a <sup>a</sup>	104 b	48 b	35 b
Deep Hollow	66 c	83 b	29 c	50 b
Thighman	134 b	139 a	105 a	154 a
Dissolved organic carbon (mg L <sup>-1</sup> )				
Beasley	12.0 b	9.4 c	10.6 a	10.2 b
Deep Hollow	13.3 b	12.2 b	9.8 a	11.8 b
Thighman	15.9 a	16.1 a	8.8 a	19.0 a
pH				
Beasley	7.13 b	6.81 c	7.27 a	6.63 a
Deep Hollow	7.30 a	7.01 b	7.11 b	6.42 b
Thighman	7.29 a	7.10 a	7.18 b	6.54 ab
EC (mmhos cm <sup>-1</sup> )				
Beasley	89 c	74 b	61 b	78 b
Deep Hollow	177 b	100 b	87 b	88 b
Thighman	282 a	242 a	206 a	251 a
Ammonium (mg L <sup>-1</sup> )				
Beasley	0.13 b	0.12 b	0.04 b	0.02 b
Deep Hollow	0.08 b	0.11 b	0.02 b	0.01 b
Thighman	0.27 a	0.17 a	0.08 a	0.05 a
Nitrate (mg L <sup>-1</sup> )				
Beasley	0.20 b	0.17 b	0.09 b	0.19 b
Deep Hollow	0.12 b	0.18 b	0.05 b	0.18 b
Thighman	0.60 a	0.32 a	0.41 a	0.94 a
Total orthophosphate (mg L <sup>-1</sup> )				
Beasley	0.89 a	0.75 ab	0.81 a	0.53 a
Deep Hollow	0.61 b	0.66 b	0.54 b	0.61 b
Thighman	0.62 b	0.82 a	0.74 a	0.99 a
Filterable orthophosphate (mg L <sup>-1</sup> )				
Beasley	0.13 a	0.18 a	0.25 a	0.08 b
Deep Hollow	0.11 a	0.12 a	0.12 b	0.06 b
Thighman	0.09 a	0.13 a	0.13 a	0.20 a

<sup>a</sup> Mean of three replicates for each sample time, means followed by the same letter do not differ at the 95% confidence level as determined by Fishers Least Significant Difference.

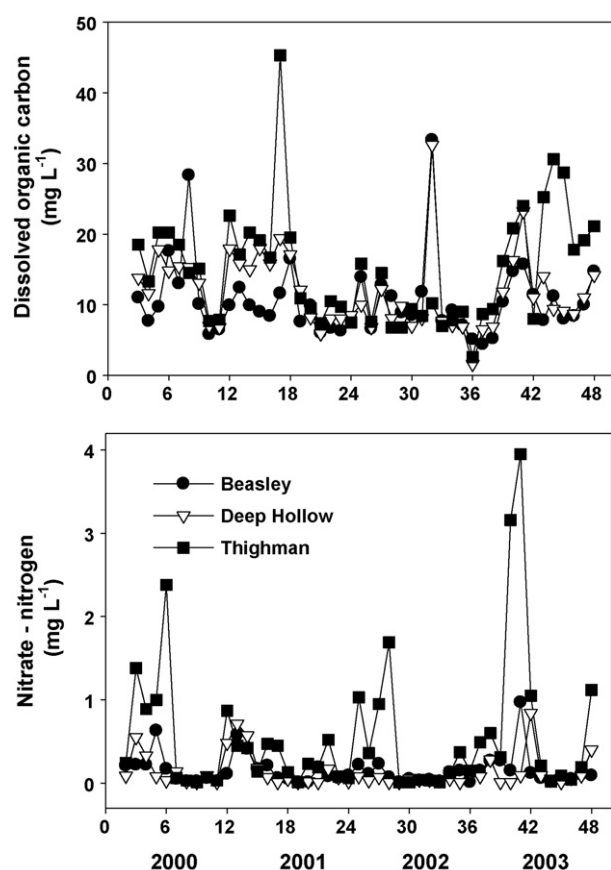


Fig. 2. Monthly total organic carbon content of three Mississippi Delta oxbow lakes, 2000–2003, each symbol represents the mean of three replicates.

in 2002 was about half that of the other 3 years. Few studies have assessed the impact of cultivation patterns in agricultural land on DOC content of lake water. In a forestry system comparing watersheds, the export of dissolved organic carbon was less in a clear cut watershed compared to a native watershed (Meyer and Tate, 1983). Aquatic DOC can arise from leaching and runoff from adjacent landscape and may also be derived from the activity and death of phytoplankton. As much as 50% of the photosynthetically fixed carbon may be exuded into the environment due to extracellular leakage and cell lysis (Fogg, 1977; Nagata, 2000). The spike in DOC observed in Beasley and Deep Hollow Lake in August of 2002 corresponds to a peak in chlorophyll *a*, lutein and  $\beta$ -carotene (Fig. 3), also diadinoxanthin and fucoxanthin (data not shown), supporting that some DOC in oxbow lakes may be arise from planktonic blooms. Greater inorganic nitrogen (ammonium and nitrate) corresponded with highest levels of DOC observed in Thighman Lake, with similar concentrations in Beasley and Deep Hollow Lakes (Table 1). In addition, 2002 had the lowest nitrate concentrations in all three lakes, while ammonium levels were lowest in 2002 and 2003.

Fluorescein diacetate (FDA) hydrolytic activity was about two-fold greater in Thighman Lake compared to Deep Hollow Lake and about four-fold higher compared to Beasley Lake (Table 2 and Fig. 4). Yearly FDA activity in both Deep Hollow and Beasley Lake was reduced in 2001–2003 compared to 2000, while Thighman Lake FDA activity remained relatively constant. Fluorescein diacetate is a generic substrate for hydrolytic enzymes such as esterases, lipases and proteases (Zablotowicz et al., 2000), consequently FDA activity can represent the total metabolic potential of a biological community and has been suggested as a tool for evaluating effects of contaminants on aquatic exposure to xenobiotics (Regel et al.,



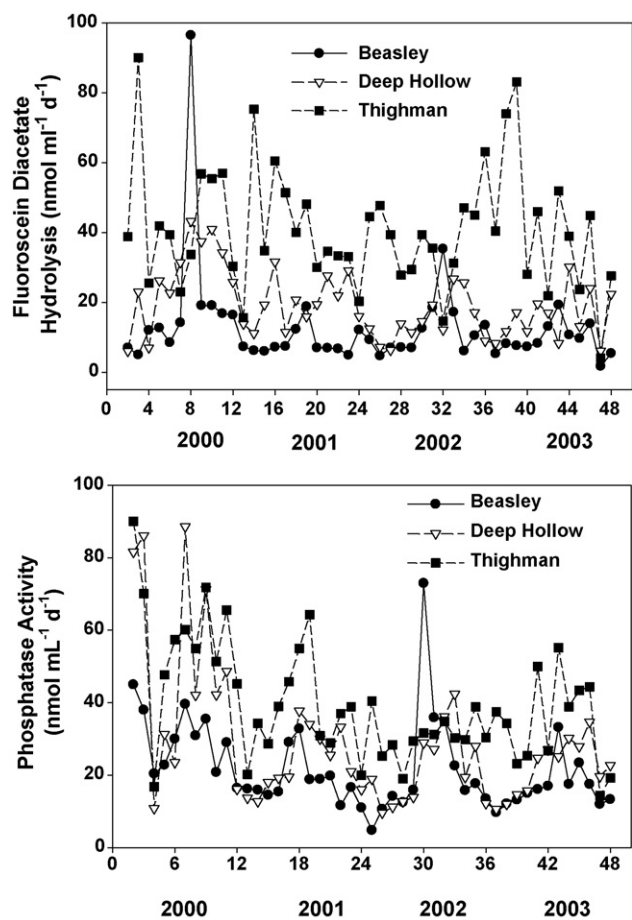


Fig. 3. Monthly means photosynthetic pigments, chlorophyll *a*, lutein, zeaxanthin, and diadinoxanthin concentration observed in three Mississippi Delta oxbow lakes, each symbol represents the mean of three replicates.

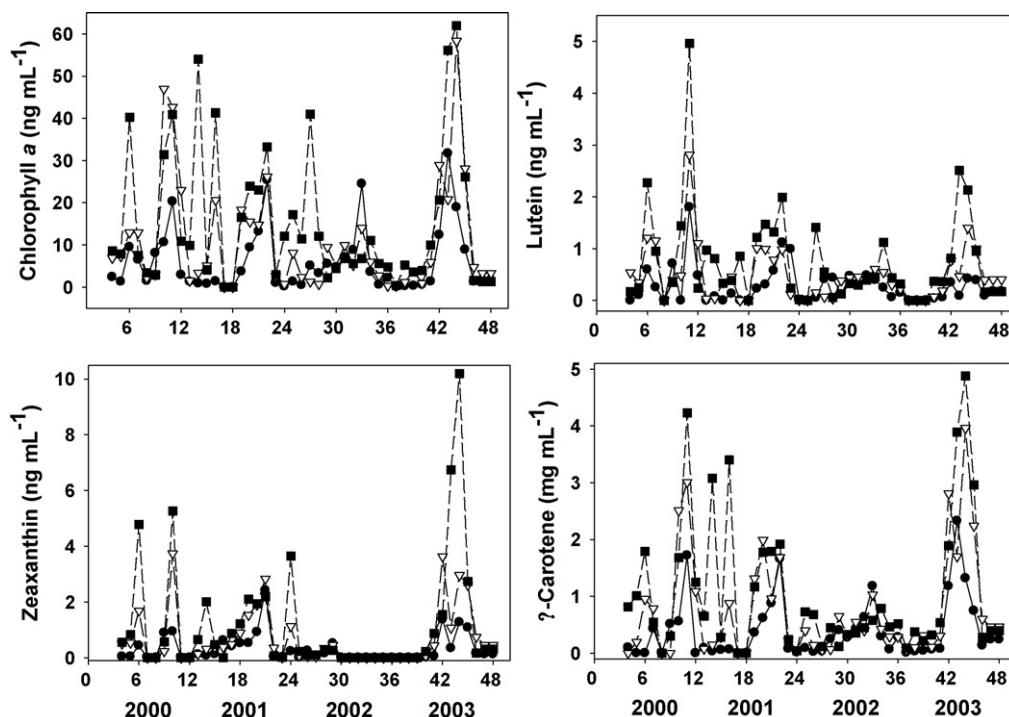


Fig. 4. Monthly means fluorescein diacetate hydrolysis and alkaline phosphatase activity of three Mississippi Delta oxbow lakes, each symbol represents the mean of three replicates.

Table 2

Yearly mean of fluorescein diacetate hydrolysis, phosphatase activity, and Biolog substrate utilization in water samples collected from three MSEA Mississippi Delta oxbow lakes (2000,  $n = 11$ ; 2002–2003,  $n = 12$ ).

Lake	2000	2001	2002	2003
FDA hydrolysis (nmol fluorescein formed mL <sup>-1</sup> 24 h <sup>-1</sup> )				
Beasley	19.7 c <sup>a</sup>	7.3 c	9.6 c	9.3 b
Deep Hollow	28.4 b	19.4 b	14.6 b	14.3 b
Thighman	38.0 a	39.7 a	37.9 a	40.4 a
Phosphatase activity (nmol p-nitrophenol formed mL <sup>-1</sup> 24 h <sup>-1</sup> )				
Beasley	29.9 c	18.4 c	23.4 b	16.7 c
Deep Hollow	49.3 b	23.3 b	21.7 b	22.0 b
Thighman	57.4 a	36.9 a	30.8 a	33.1 a
Biolog substrate utilization, average well density (od 585 nm)				
Beasley	0.91 a	1.08 b	1.10 b	0.83 b
Deep Hollow	1.05 a	1.40 a	0.89 b	0.77 b
Thighman	0.91 a	1.30 a	1.34 a	1.23 a

<sup>a</sup> Mean of three replicates for each sample time, means followed by the same letter do not differ at the 95% confidence level as determined by Fishers Least Significant Difference.

2002; Gilbert et al., 1992) and assessing nutrient status of phytoplankton (Brookes et al., 2000). The higher FDA activity in Thighman Lake is consistent with higher nutrient availability as suggested by levels of DOC and EC to support bacterial heterotrophic activity. Alkaline phosphatase activity was highest for all lakes in 2000 and declined thereafter, with a pattern of the greatest activity in Thighman Lake, intermediate in Deep Hollow and lowest in Beasley Lake (Table 2 and Fig. 3). The overall decrease in alkaline phosphatase activity in Thighman Lake corresponded to increasing levels of FOP. However, levels of FOP decreased in Deep Hollow Lake with decreasing alkaline phosphatase activity. Phosphate availability can regulate expression of alkaline phosphatase activity in bacterioplankton and phytoplankton, however, similar relationships between FOP and enzyme activities were not observed in this study. No concrete pattern of seasonality of activity was observed and in some cases there was a spike in phosphatase activity that corresponded to a peak in DOC (e.g. Beasley Lake August 2002). As in the

case of FDA activity, no pattern of phosphatase activity was associated with changes in watershed activity in either Beasley or Deep Hollow.

Substrate utilization assays (Biolog plates) indicate a similar carbon utilization potential among all lakes in 2000 (Table 2). However in 2001–2003 significant differences in carbon utilization were observed among all lakes with the greatest potential in Thighman Lake and lowest in water samples from Beasley Lake. In 2001 water from Deep Hollow Lake had a similar level of substrate utilization as Thighman Lake and declined in the two subsequent years. This change can indicate the shift in the bacterioplankton population during the transition from a no-till watershed to conventional tillage managed system. Although the BIOLOG system has been used to characterize patterns of functional succession in studies of diversity of bacterioplankton (Matsui et al., 2001), the response of the water samples was not normalized to a cell density and therefore was used solely to characterize metabolic potential. In addition the range of substrates used by most water samples was similar with at least 70–85 of the 95 substrates utilized in every assay. Most carbohydrates and L-amino acids were readily used. Substrates with infrequent use include the carbohydrates cellobiose,  $\beta$ -methyl glucoside, and psicose, the organic acids  $\gamma$ -hydroxybutyrate,  $\alpha$ -ketobutyric acid, succinamic acid, and the amino acids hydroxyl-L-proline, and the amines 2-amino ethanol and phenylethylamine.

### 3.2. Bacterioplankton dynamics

The yearly mean of various bacterial components of the three lakes are presented in Table 3. As previously reported elsewhere (Zablotowicz et al., 2004) total and Gram-negative bacterial populations were most abundant in Thighman Lake. Following a reversion of Deep Hollow watershed to conventional tillage in 2001, bacterial populations increased to a similar density as Thighman Lake water, but thereafter declined to levels similar to Beasley Lake. The highest abundance of bacterioplankton in Thighman Lake co-occurs with the highest DOC and biochemical indicators (FDA hydrolysis and alkaline phosphatase activity). Coliform estimates ranged from a low of  $\sim \log 1.0$ – $2.6$  cells mL<sup>-1</sup> with populations observed in Deep Hollow typically the lowest. Beasley watershed has the greatest area in riparian forest (135 ha) that provides habitat for fauna and potentially serves as a source for this group of bacteria. Thighman watershed is unique among the three watersheds in that it has a significant amount of catfish ponds in and surrounding the watershed (18% of the watershed area). There has been limited evidence for drainage or seepage of water from the ponds into the lake, and

**Table 3**

Mean cultural heterotrophic bacterioplankton populations of water samples collected from three MSEA Mississippi Delta oxbow lakes (2000,  $n = 11$ ; 2002–2003,  $n = 12$ ).

Lake	2000	2001	2002	2003
Total heterotrophic bacteria (log [10] colony forming units mL <sup>-1</sup> )				
Beasley	4.08 b <sup>a</sup>	4.54 b	4.36 b	4.04 b
Deep Hollow	4.19 b	5.14 a	4.18 c	3.91 b
Thighman	4.66 a	5.28 a	5.07 a	4.69 a
Gram-negative bacteria (log [10] colony forming units mL <sup>-1</sup> )				
Beasley	3.17 b	3.18 c	3.05 c	2.78 b
Deep Hollow	3.25 b	4.10 b	3.44 b	2.59 b
Thighman	3.64 a	4.31 a	3.86 a	3.35 a
Coliforms (log [10] cells mL <sup>-1</sup> )				
Beasley	1.44 a	1.88 b	2.14 b	0.99 b
Deep Hollow	1.01 b	1.89 b	1.26 c	0.99 b
Thighman	1.69 a	2.62 a	2.49 a	1.82 a

<sup>a</sup> Mean of three replicates for each sample time, means followed by the same letter do not differ at the 95% confidence level as determined by Fishers Least Significant Difference.

**Table 4**

Yearly mean algal populations as determined by MPN techniques of water samples collected from three MSEA Mississippi Delta oxbow lakes (2000,  $n = 11$ ; 2002–2003,  $n = 12$ ).

Lake	log [10] propagules mL <sup>-1</sup>			
	2000	2001	2002	2003
Beasley	3.19 b <sup>a</sup>	3.25 b	2.90 c	3.55 c
Deep Hollow	3.98 a	3.69 a	3.49 b	3.88 b
Thighman	3.77 a	3.77 a	3.88 a	4.53 a

<sup>a</sup> Mean of three replicates for each sample time, means followed by the same letter do not differ at the 95% confidence level as determined by Fishers Least Significant Difference.

previous research indicates that coliform levels in catfish production ponds are typical of adjoining lakes (Boyd and Tanner, 1998). Previous reports (Knight and Welch, 2004) indicated that before imposition of BMPs, Thighman had the highest abundance of coliforms, Deep Hollow Lake was intermediate, and Beasley Lake the lowest.

### 3.3. Phytoplankton dynamics

Estimates of total algae based upon MPN methodology are summarized in Table 4 and selected photosynthetic pigment analyses are presented in Fig. 3 and Table 5. Based on MPN estimates, algal abundance was greater in Thighman Lake and Deep Hollow Lake compared to Beasley Lake in 2000 and 2001 (Table 5). However, in 2002 and 2003 estimates of populations were Thighman > Deep Hollow > Beasley Lake. Although suspended solid levels in Beasley Lake declined there was no effect on estimates of total algal propagules. Based on analysis of carotenoid and photosynthetic pigments there was a drastic reduction in the algal community in 2002, which recovered in 2003 (Table 4). The low level of pigments in 2002 coincides with lowest levels of DOC, agreeing with the hypothesis that the major source of DOC is from phytoplankton. Generally, most pigments were lowest in Beasley Lake, highest in Thighman and intermediate in Deep Hollow Lake. Chlorophyll *a* concentration

**Table 5**

Mean concentration of various photosynthetic pigments (ng mL<sup>-1</sup>) observed in three Mississippi Delta oxbow lakes from 2000 to 2003 (2000,  $n = 11$ ; 2002–2003,  $n = 12$ ).

Lake	2000	2001	2002	2003
Chlorophyll <i>a</i> (total algae)				
Beasley	7.72 b <sup>a</sup>	4.78 b	5.05 a	6.59 b
Deep Hollow	18.80 a	10.46 b	5.70 a	13.30 ab
Thighman	21.80 a	27.48 a	7.67 a	16.20 a
$\beta$ -Carotene (total algae)				
Beasley	0.41 b	0.32 b	0.30 b	1.09 a
Deep Hollow	1.07 ab	0.67 ab	0.38 ab	1.09 a
Thighman	1.45 a	1.24 a	0.47 a	1.37 a
Lutein (green algae)				
Beasley	0.49 b	0.21 b	0.26 a	0.15 b
Deep Hollow	1.00 ab	0.42 b	0.30 a	0.42 a
Thighman	1.32 a	1.40 a	0.46 a	0.60 a
Zeaxanthin (cyanobacteria)				
Beasley	0.29 b	0.49 b	0.11 a	0.38 c
Deep Hollow	0.84 ab	0.42 b	0.17 a	1.03 b
Thighman	1.62 a	1.21 a	0.36 a	1.95 a
Diadinoxanthin (diatoms)				
Beasley	0.33 b	0.13 b	0.44 a	0.28 b
Deep Hollow	1.99 a	0.81 a	0.47 a	0.98 a
Thighman	1.47 a	0.89 a	0.36 a	0.85 a
Fucoxanthin (diatoms and dinoflagellates)				
Beasley	0.22 b	0.30 a	0.25 a	0.10 b
Deep Hollow	0.72 a	0.70 a	0.46 a	0.42 a
Thighman	0.67 a	0.66 a	0.50 a	0.42 a

<sup>a</sup> Mean of three replicates for each sample time, means followed by the same letter do not differ at the 95% confidence level as determined by Fishers Least Significant Difference.

was high and remained uniform suggesting the carrying capacity for these lakes was attained, except in 2002. Diatoms (using diadinoxanthin carotenoid) were most abundance in spring and late fall. Green algae (all of which contain lutein) had a summer and early winter maxima. Cyanobacteria (all contain zeaxanthin) were maximal in late summer with notably low abundance in 2002, and levels increased for all lakes again in 2003. Drastic changes in zeaxanthin may indicate changes in N availability causing a shift to/from filamentous to coccoid species. However the same phenomena occurring in all three lakes suggest that other environmental factors may have limited cyanobacterial abundance in 2002.

### 3.4. Canonical discriminant analysis

A summary of canonical discriminant analysis for the three lakes analyzed by year is presented in Fig. 5. In all cases CAN1 and CAN2 accounted for 84–93% of the variability, and the model significance was  $>0.0001$ . In all years, CAN1 axis accounted for over 73–95% of variation in the explanatory model, and in all cases CAN1 and CAN2 accounted for  $>99\%$  of the lake separation, and the model significance was  $p < 0.0001$ . During year 2 (2001) the relative importance of CAN1 was less than other years (70+%), with the CAN2 axis accounting for a much higher percentage of the model variation than other years. Electrical conductivity was the most important variable contributing to the CAN1 axis in all 4 years. The CAN1 axis was also highly affected by FDA activity in all years except 2001, when total solids impacted the CAN1 axis. This year was the transition year between management practices. Thigman watershed had the least change in management practices and there were few changes in measured variables. Thigman Lake consistently was separated from the other two lakes, especially as Thigman had the highest EC, solids, DOC and most biological parameters. Beasley Lake initially had the highest solids, these values decreased by 75% relative to starting conditions. Deep Hollow had a large decrease in

EC and subsequent to the reversion to conventional tillage both FDA and phosphatase activity decreased. Considering bacterioplanktonic and phytoplanktonic parameters that contributed to the CANDISC model, Chlorophyll *a* was more important in accounting for model separation on CAN1 axis in 2001 compared to years 2002 and 2003, while total bacteria levels were a major contributor in 2002. In 2000, 2002 and 2003, CAN2 represented several algal pigments including  $\beta$ -carotene, lutein and chlorophyll *b*, these are indicative of green algae. In 2002 and 2003 zeaxanthin was a major component of CAN2 separation, and was negatively associated in 2002, whereas in 2003 it was the strongest positive contributor in agreement with observed pigment values. The diversity and distribution of certain cyanobacterial species has been found to be most widespread in freshwater systems with low conductivity (Hamed, 2008), although many species are tolerant to quite a range of specific ion conditions. The abundance of cyanobacteria in Spanish rice fields was positively correlated with conductivity, reactive phosphate, and salinity, while dissolved inorganic nitrogen was negatively correlated with cyanobacterial abundance (Quesada and Fernández-Valiente, 1996). The diatoms, however, are more sensitive to conductivity and specific ions (Hamed, 2008; Reid, 2005).

A summary of canonical discriminant analysis for the yearly structure of the three individual lakes is presented in Fig. 6. In this analysis, CAN1 axis accounted for 53–64% of variation in the explanatory model, and CAN2 accounted for 20–32% of the yearly separation, with a model significance of  $p < 0.0001$  for CAN1 and CAN2. Of the chemical parameters, EC was a positive contributor to yearly separation in CAN1 in Beasley Lake and Deep Hollow Lake, positive in CAN2 in Beasley Lake and negative in CAN2 in Thigman Lake. Suspended solids were a dominant contributor to canonical structure only in Beasley Lake (negative in CAN1 and positive in CAN2). FDA-hydrolytic activity was positively associated with both CAN1 and CAN2 in Thigman Lake, and negatively associated with CAN1 in Deep Hollow Lake, however, phosphatase

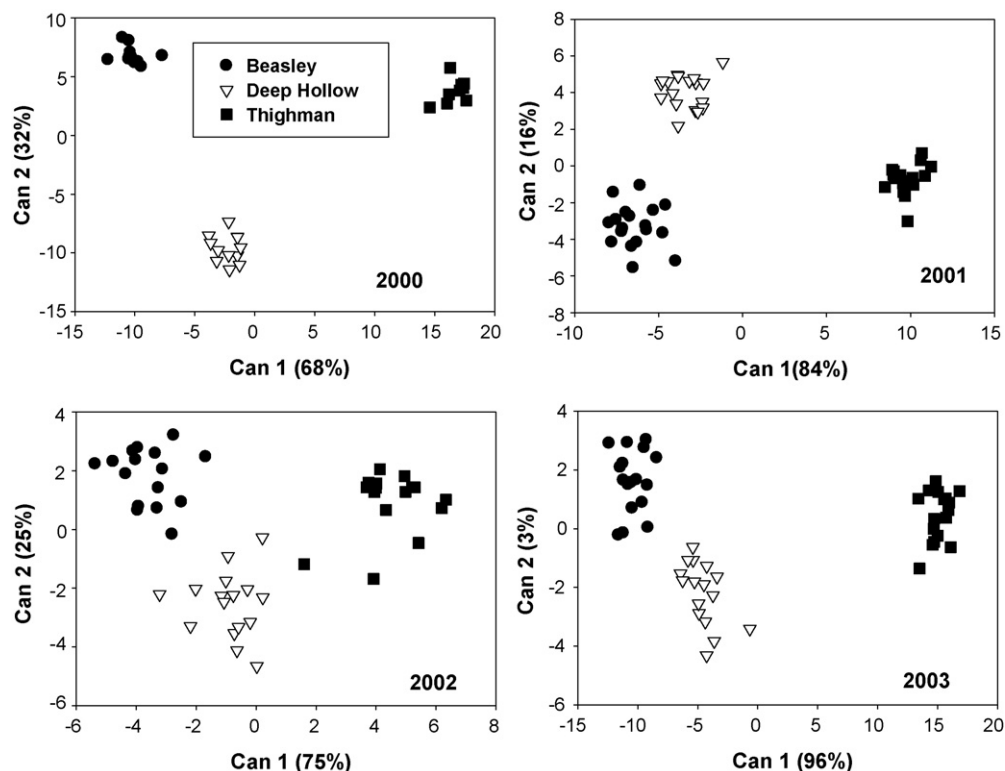
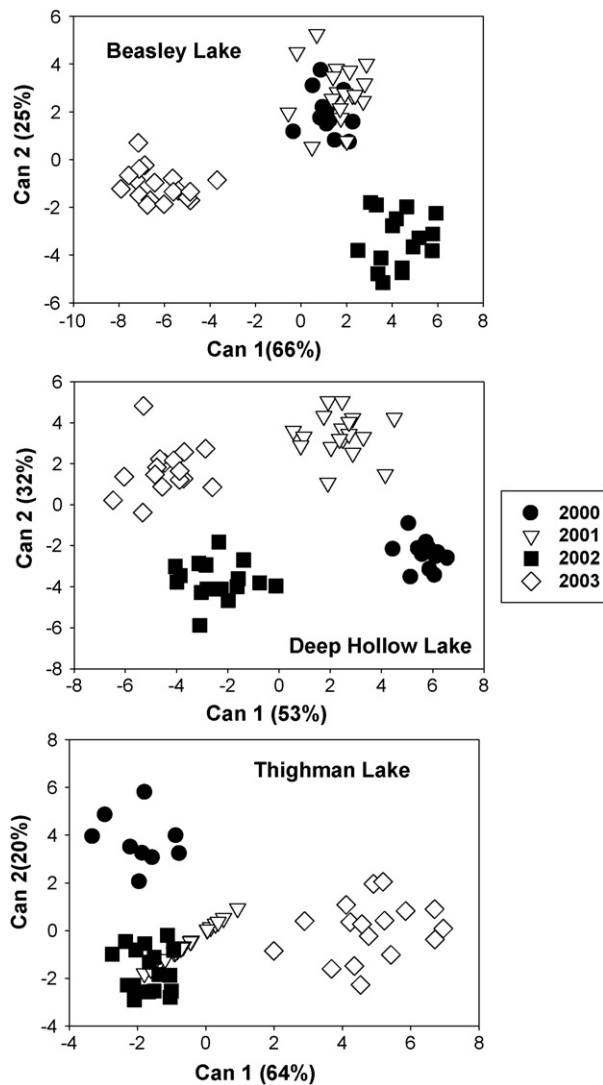


Fig. 5. Plots of canonical discriminant analysis assessing yearly changes in chemical, physical, biochemical and biological parameters from the three Mississippi Delta oxbow lakes during years 2000–2003.



**Fig. 6.** Plots of canonical discriminant analysis assessing fluctuations of chemical, physical, biochemical and biological parameters from the 3 years plotted against year.

activity contributed to CAN1 structure in Deep Hollow Lake only. Of the pigments chlorophyll *a* was negatively contributed to CAN1 in Thighman Lake and negatively contributed to CAN2 in all three lakes, while zeaxanthin contributed to CAN2 in Both Deep Hollow and Thighman Lake. Several other diagnostic pigments  $\beta$ -carotene, lutein, fucoxanthin, and myxoxanthin contributed to the canonical structure of the three individual lakes. In regards to bacterial groups, total bacteria negatively contributed to both CAN1 in Thighman Lake and negatively contributed to CAN2 in Deep Hollow Lake and Thighman Lake. These analyses indicate which unique factors contribute to the yearly variation of the three individual lakes.

There are two significant time dependent changes in canonical structure in Beasley and Thighman lakes while Deep Hollow remained relatively unchanged. In Beasley watershed, conservation tillage was implemented in 2002 with the adoption of a majority of the arable land planted in glyphosate-resistant crops, especially soybeans (Zablotowicz et al., 2006; Locke et al., 2008). An additional contributed to decreases in suspended solids during 2002 and 2003 is decreased rainfall amount for those years (Zablotowicz et al., 2006). Associated with the adoption of reduced tillage was a significant decrease in suspended solids which was a significant component of both CAN1 and CAN2 in Beasley Lake. Within Thighman Lake, major changes in CAN structure were observed in 2003 along CAN1 compared to previous 3 years. In Thighman Lake, biological properties such as bacterial populations and enzymatic activity and photosynthetic pigments were the major contributing factors to canonical discrimination, especially in CAN1. Only two physical components of water quality (temperature and electrical conductivity; Table 6) contributed to CAN2 for this watershed. During winter of 2003, average water temperatures were about 5 °C warmer than previous years and as result may have stimulated biological parameters. In addition, a significant increase in heterotrophic activity (as FDA hydrolysis) occurred during the winter and spring of 2003 (Fig. 4a). In Deep Hollow watershed, 2000 was the last year for conservation tillage practices and thereafter CAN1 ordinates moved to the negative position. In 2000 the greatest contributing factors were physical and chemical parameters (FOP and EC; Table 7). In the shift from 2000 to 2003, there was a decrease in both parameters of enzymatic activity (FDA hydrolysis and alkaline phosphatase) however observed changes in water quality were difficult to ascertain with changes in land-use management practices in Deep Hollow watershed. Previous research in Deep Hollow watershed conducted from 1997 to 1999 showed 76% reduction in suspended solids and a 150% increase in chlorophyll concentrations (Cullum et al., 2006) associated with implementa-

**Table 6**

Dominant biological, chemical and physical parameters contributing to canonical structure of the three oxbow lakes.

Lake	Direction	CAN1	CAN2
Beasley	Positive	pH Electrical conductivity	Electrical conductivity Solids Algae (MPN)
	Negative	Myxoxanthin Solids	Temperature Fucoxanthin Chlorophyll <i>a</i>
Deep Hollow	Positive	Electrical conductivity Coliforms Lutein	Zeaxanthin $\beta$ -Carotene Total bacteria
	Negative	Phosphatase activity Fluorescein diacetate hydrolysis $\beta$ -Carotene	Chlorophyll <i>a</i> pH
Thighman	Positive	$\beta$ -Carotene Fluorescein diacetate hydrolysis Gram-negative bacteria	Fluorescein diacetate hydrolysis Zeaxanthin Temperature
	Negative	Chlorophyll <i>a</i> Total bacteria	Total bacteria Electrical conductivity Chlorophyll <i>a</i>



**Table 7**

Dominant biological, chemical and physical parameters contributing to canonical structure of the three oxbow lakes on a yearly basis.

Year	Direction	CAN1	CAN2
2000	Positive	Filterable orthophosphate Total bacteria Electrical conductivity	Total bacteria Zeaxanthin
	Negative	Total orthophosphate Zeaxanthin Gram-negative bacteria	Filterable orthophosphate
2001	Positive	Electrical conductivity Lutein	FDA-hydrolytic activity Biolog
	Negative	Chlorophyll <i>a</i> Filterable orthophosphate	Lutein Temperature
2002	Positive	Zeaxanthin Electrical conductivity Phosphatase activity	Nitrate Temperature
	Negative	Temperature Chlorophyll <i>a</i> Dissolved oxygen	Zeaxanthin Algae MPN
2003	Positive	Electrical conductivity FDA-hydrolytic activity Zeaxanthin	
	Negative	Fucoxanthin Dissolved oxygen Temperature Chlorophyll <i>a</i>	

tion of conservation tillage and edge-of-field practices. In contrast, the current study showed limited change in physical water quality measurements associated with changes from conservation tillage back to conventional tillage from 2001 to 2003.

#### 4. Conclusions

Although there is considerable variation in physical, chemical, biochemical, and biological parameters measured in this study of three oxbow lakes that can be affected by seasonal and yearly differences in environmental conditions, some general observations can describe the effects of land management practices on water quality and the biological properties of surface water. There were three chemical and physical parameters that contributed to the biological activity and subsequent bacterioplankton and phytoplankton population dynamics in oxbow lakes: suspended solids, electrical conductivity and DOC. Conductivity is directly related to alkalinity (carbonate concentration), which is well known to regulate aquatic production. Dissolved organic carbon serves as a limiting nutrient for bacteria in a similar fashion (Wetzel and Likens, 1995). Fluorescein diacetate hydrolytic activity is a suitable tool for predicting heterotrophic biochemical activity in oxbow lakes. Canonical analysis proved useful for analyzing a range of parameters in quantifying changes in lake ecology resulting from changes in agricultural landscape management.

Future studies should involve more rigorous sampling both spatially and temporally. Phytoplankton patch size can vary on scales of meters, with temporal differences on scales of minutes to hours (Zimba and Gitelson, 2006). We would recommend more intensive sampling in both spatial and temporal scales to better model patterns observed in this study. In addition, Phase III of the MD-MSEA project has been redirected into the Conservation Effects Assessment Project (CEAP) with more intensive conservation practices emplaced into Beasley watershed. Transition of Beasley watershed to a national CEAP watershed within the Lower Mississippi River Basin accentuates the need for expanded research to help address issues such as Gulf Hypoxia.

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